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REGULAR ARTICLE

BIOACTIVE COMPOUNDS IN COMMONLY UTILIZED LEGUME CULTIVARS FROM IRAQ

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ABSTRACT

Total polyphenol content as well as total antioxidant capacity in three chickpea, four fababean and four lentil cultivars from regions of Iraq with different environmental growth conditions were investigated in the work. The total polyphenol content (TP) was estimated using Folin-Ciocalteau assay and the total antioxidant capacity (TAC) of legume extracts was measured using the DPPH spectrophotometrically. The obtained results confirmed that the polyphenols content in the tested legume cultivars was influenced by locality. From tested legumes the highest polyphenol content was measured in fababean (1220 – 6286 mg GAE.kg^{-1}). In lentil (2351 – 3011 mg GAE.kg^{-1}) the average TP value was slightly lower (by 1,6%), while in chickpea (549 – 978 mg GAE.kg^{-1}) it was dramatically (by 71,5%) lower in comparison to fababean. The similar trend was observed at values of TAC. The highest average TAC value was determined in fababean (15.2 – 25.6% DPPH), in lentil (14.2 – 28.9% DPPH) the TAC value was by 2% and in chickpea (1.9 – 4.5% DPPH) by 83,6% lower in comparison to fababean. Only in lentil a statistically strong correlation (*P-value* 2.391E-06; R = 0.802) was found. Our results confirmed that legumes can be a good source of bioactive compounds in the human nutrition.

Keywords: fababean, chickpea, lentil, total polyphenol content, total antioxidant capacity

INTRODUCTION

Legume grains have been playing a key role in the traditional diets of human beings throughout the world. They are excellent source of protein, dietary fiber, starch, micronutrients and bioactive compounds with low level of fat (**Chang *et al.*, 2000**). The bioactive principles are divided chemically into a number of groups among which are alkaloids, volatile essential oils, phenols and phenolic glycosides, resins, oleosins, steroids, tannins and terpenoids (**Ferreira *et al.*, 2008**), antioxidants (**Heimler *et al.*, 2005**) and anticarcinogenic compounds (**Hangen, Bennink, 2002; Mishra *et al.*, 2010**). Evaluation of antioxidant activity of phenolic compounds from leguminous seeds has been of interest in recent year. Phenolic compounds, such as phenolic acids, flavonols, flavones, isoflavones, anthocyannins, and condensed tannins, have been identified and characterized in food legumes (**Beninger and Hosfield, 2003; Madhujith *et al.*, 2004; Xu *et al.*, 2007a, b**). Several studies have reported on antioxidant and antiradical activity of tannins (**Troszyńska and Kubicka, 2001; Amarowicz *et al.*, 2004; Alasalvar *et al.*, 2006; Amarowicz, 2007**).

The aim of the work was to evaluate the content of bioactive compounds (polyphenolics) in legumes commonly utilized in the human diet in Iraq, to compare their antioxidant capacity and to evaluate the influence of grown locality on observed parameters.

MATERIAL AND METHODS

Three chickpea, four fababean and four lentil cultivars were cultivated in fields of regions in Iraq with different environmental growth conditions.

Extraction

For 12 hours extraction, dry material (5 g) was used and continuously extracted by a Twisselmann extractor with methanol (80%, v/v).

Total polyphenols

Total polyphenols were determined by the method of **Lachman *et al.* (1998)** and expressed in mg eq. gallic acid per kg dry matter. The total polyphenol content was estimated using Folin-Ciocalteau assay on the spectrophotometer Shimadzu 710 (Japan).

Antioxidant aktivity

The free radical scavenging activity of the extracts was measured using the DPPH (1,1-diphenyl-2-picrylhydrazyl) method of **Brand-Williams *et al.* (1995)** spectrophotometrically (Shimadzu UV/VIS-1240).

Statistical analysis

Statistical processing of the results was carried out Statistica 8. One-way ANOVA was used. Mean comparisons between cultivars were done by the LSD test.

RESULTS AND DISCUSSION

According to the obtained results, the polyphenols content (TP) in the tested cultivars was significantly different and was influenced by locality.

Table 1 Total polyphenols content (mg eq. gallic acid.kg^{-1} DM) and antioxidant activity (% of DPPH inhibition) in chickpea cultivars

cultivar (locality)	TP	AOA
C1 (Marakeshi)	555.12±4.13 a	4.01±0.33 b
C2 (Erbil Research)	969.30±6.39 c	3.87±0.24 a,b
C3 (Harer)	770.68±19.92 b	2.08±0.11 a

Data expressed as means of six replications ± standard deviation. Values in the same column with the different letters present significant differences using t-test for independent samples.

Our results suggest that severe climatic conditions have caused a slight increase in the total content of polyphenolic substances. For growing chickpeas are the best warm and dry areas. Therefore the C1 seeds have shown significantly much lower TP content (this cultivar is grown in area suitable for growing chickpea), while the C3 variety, grown in mountain areas with higher rainfall, recorded higher amount of polyphenols. **Lachman *et al.* (2006)** investigated the effect of weather conditions on the TP contents of potatoes and their results showed that an upland cooler site with higher rainfall provide the tubers with a higher content of TP. On the other hand, the highest TP value of cv. C2 indicate influence of cultivar on TP content, too.

About the impact site is not enough literature demonstrable knowledge and scientists attache greater importance to other factors: the influence of variety, vintage effects and stress factors. Antioxidant capacity was genotype dependent, ranging from 2.08 to 4.01% of DPPH inhibition, but unaffected by location.

Table 2 Total polyphenols content (mg eq. gallic acid.kg^{-1} DM) and antioxidant activity (% of DPPH inhibition) in fababean cultivars

cultivar (locality)	TP	AOA
F1 (Aqra)	1898.71±26.20 a	16.62±0.81 a
F2 (Erbil, Ankawa)	2702.44±37.95 a,b	24.54±0.64 d
F3 (Alton kupri)	1583.54±13.79 b	21.66±2.12 b
F4 (Erbil Research)	3374.91±13.38 a	18.24±0.39 c

Data are expressed as means of six replications ± standard deviation. Values in the same column with the different letters present significant differences $p < 0.05$ using t-test for independent samples.

Our results suggest that more severe climatic conditions have caused a slight increase in the total content of polyphenolic substances (Table 2). For growing fababean warm and dry areas appears as the best. Therefore the F3 seeds from Alton kupri locality have shown the lowest TP content (this cultivar is grown in area suitable for growing fababean), while in the same F1 cultivar grown in mountain area (Aqra) with higher rainfall, was recorded more higher TP amount. **Lachman *et al.* (2006)** investigated the effect of weather conditions on the TP contents of potatoes and their results also showed that an upland cooler site with higher rainfall provide the seeds with a higher content of TP. On the other hand, the highest TP value of cv. F2 or F4 from Erbil locality with medium rainfall also indicates more influence on TP content.

Antioxidant capacity was in relation to genotype, ranging from 16.62 to 24.54 % of DPPH inhibition, but unaffected by location in varieties grown in Iraq.

About the impact site is not enough literature demonstrable knowledge and scientists attaching greater importance to other factors: the influence of variety, and stress factors.

Table 3 Total polyphenols content (mg eq. gallic acid.kg^{-1} DM) and antioxidant activity (% of DPPH inhibition) in lentil cultivars

cultivar (locality)	TP	AOA
L1 Qushtopa	2863.60±95.82 d	25,85±2,09 c
L2 Shaklawa	2419.62±47.99 a	16,66±1,56 a
L3 Kalak	2762.07±60.25 c	21,07±1,05 b
L4 Koysinjaq	2533.01±81.27 b	15,85±0,40 a

Data are expressed as means of six replications ± standard deviation. Values in the same column with the different letters present significant differences $p < 0.05$ using t-test for independent samples.

The determined TP content in lentil was in interval 2351 – 3011 mg GAE.kg^{-1}, the determined TAC value in interval 14.2 – 28.9% DPPH (Table 3). **Gillooly (1983)** presented higher values of the total polyphenol content in brown and green lentils (6800 mg.kg^{-1} and 6300 mg.kg^{-1} respectively). **Awada *et al.* (2005)** determined lower polyphenol content in lentil seeds (4730 mg.kg^{-1}) in comparison to mentioned authors. On other hand the values of total polyphenols in lentil presented by **Sindhu *et al.* (2012)** (1191 mg.kg^{-1}) are significantly lower than our results. The obtained results confirmed that the polyphenols content in the tested lentil cultivars was influenced by locality.

Along with essential nutrients, lentils are good sources of many nonnutrient functional phytochemicals such as phytic acid and tannins **(Vidal-Valverde *et al.*, 1994)**, which are considered among the functional antioxidant ingredients **(Scalbert *et al.*, 2005; Vucenik, Shamsuddin, 2006)**. Lentils are also good sources of the antioxidants catechins and procyanidins **(Auger *et al.*, 2004)**. As a consequence of the presence of a wide spectrum of phenolic and nonphenolic antioxidants in lentils, they have shown the highest antioxidant and oxygen radical-absorbing capacities among different legumes **(Xu and Chang, 2007, 2008)**. Natural phenolic antioxidants can scavenge reactive oxygen and nitrogen species (RONS) thereby preventing of oxidative diseases in the body **(Willett, 1994)**.

From tested legumes the highest polyphenol content was measured in fababean. In lentil the average TP value was slightly lower (by 1.6%), while in chickpea it was dramatically (by 71,5%) lower in comparison to fababean (Figure 1). The similar trend was observed at values of TAC (Figure 2). The highest average TAC value was determined in fababean, in lentil the TAC value was by 2% and in chickpea by 83,6% lower in comparison to fababean.

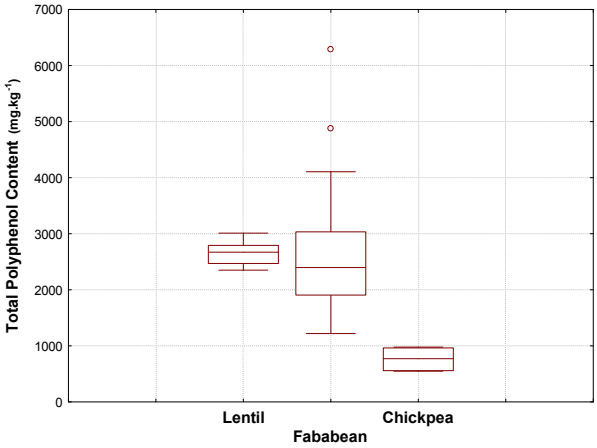

Figure 1 Polyphenol content in tested legumes (mg GAE.kg^{-1})

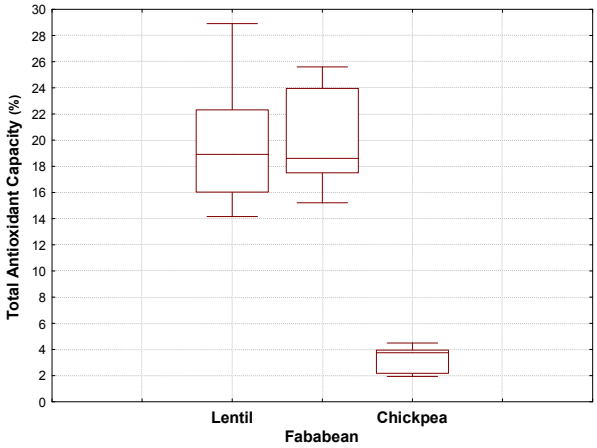

Figure 2 Total antioxidant capacity in tested legumes (% DPPH)

Neither in fababean (*P-value* 0.797, Figure 3) nor in chickpea (*P-value* 0.767, Figure 4) were confirmed correlations between total polyphenol content and total antioxidant capacity values. Only in lentil a statistically strongly significant correlation (*P-value* 2.391E-06; R = 0.802) was found (Figure 5). **Amarowicz *et al.* (2005)** analysed the extracts of fababean, broad bean, adzuki bean, red bean, pea, red lentil and green lentil seeds using 80% (v/v) acetone and confirmed a statistically significant correlation between the total antioxidant activity values and total phenolics (P = 0.01). A strong correlation between total polyphenol content and antioxidant activity (R = 0.86; P<0.05) was observed also by **Akond *et al.* (2011)** in common bean. According these authors this finding suggests that total polyphenol content is a good predictor of in vitro antioxidant activity.

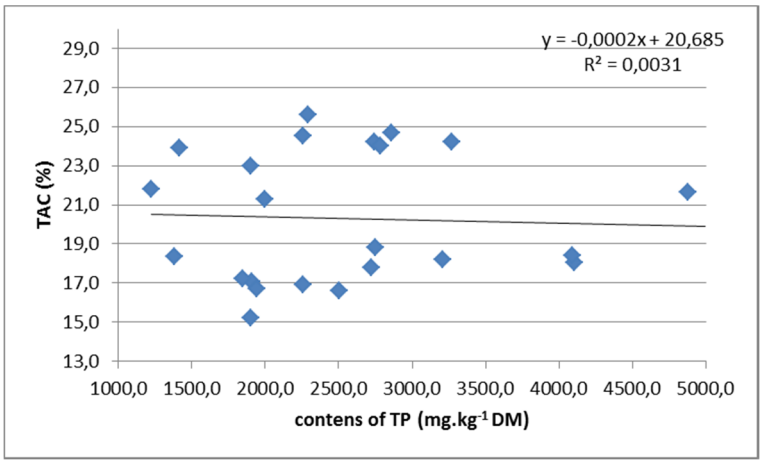

Figure 3 Correlation between TP and TAC (fababean)

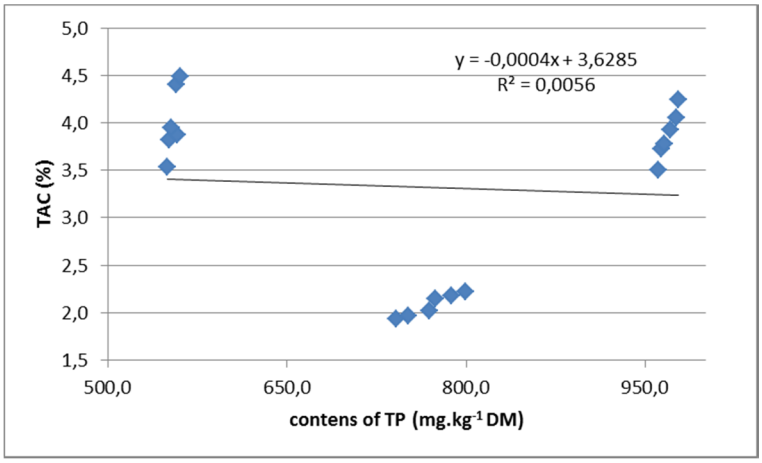

Figure 4 Correlation between TP and TAC (chickpea)

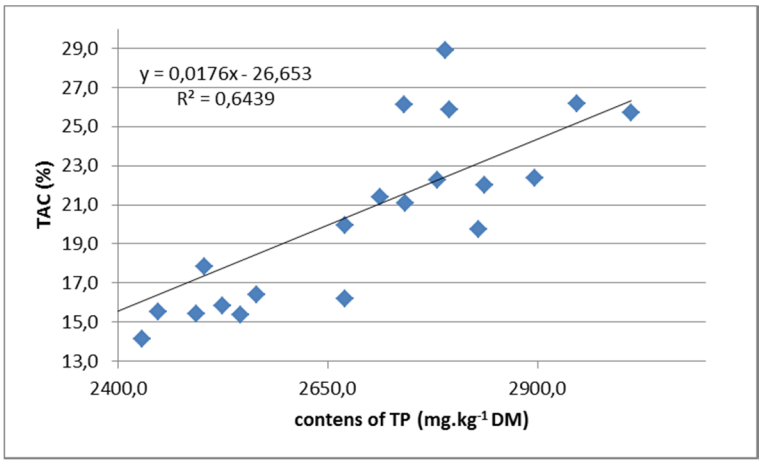

Figure 5 Correlation between TP and TAC (lentil)

CONCLUSION

Fababean, chickpea and lentil belong to commonly utilized legumes in Iraq and they play a key role in the traditional human diet in this country. Based on our results the followed order of total polyphenol content as well as total antioxidant capacity in investigated legumes obtained from Iraq can be created: fababean > lentil > chickpea. Our results confirmed that legumes can be a good source of bioactive compounds in the human nutrition.

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